Message

From: Brasaemle, Karla [Karla.Brasaemle@TechLawInc.com]

Sent: 7/17/2020 4:17:44 PM

To: Nicole Goers [Nicole.Goers@TechLawInc.com]; D'Almeida, Carolyn [dAlmeida.Carolyn@epa.gov]

Subject: RE: Some initial thoughts on Final SIP Reports from Microbial Insights

I'm available:

August 3: After 11 am August 4: Any time August 7: Any time

Karla Brasaemle, P.G., TechLaw 415-762-0566

From: Goers, Nicole < Nicole.Goers@TechLawInc.com>

Sent: Friday, July 17, 2020 8:49 AM

To: 'D'Almeida, Carolyn' <dAlmeida.Carolyn@epa.gov> **Cc:** Brasaemle, Karla <Karla.Brasaemle@TechLawInc.com>

Subject: RE: Some initial thoughts on Final SIP Reports from Microbial Insights

I received the Microbial Insights documents.

I have a 8-9am PT call on the 4th and a 9-10am PT call on the 6th. I'm otherwise free to discuss.

Nicole

540-836-0420

From: D'Almeida, Carolyn < dAlmeida.Carolyn@epa.gov>

Sent: Friday, July 17, 2020 10:42 AM

To: Brasaemle, Karla < "> Goers, Nicole < Nicole <a hre

Subject: FW: Some initial thoughts on Final SIP Reports from Microbial Insights

Karla and Nicole

Did you receive the results of the ST12 microbial data discussed on yesterday's BCT call? See comments from Eleanor below. I'd like to schedule an internal team call to discuss with ADEQ consultants prior to the next BCT. I'll send you a TDM once we get it on the calendar. What is your availability during week of Aug 3?

Carolyn d'Almeida Remedial Project Manager Federal Facilities Branch (SFD 8-1) US EPA Region 9 Laboratory 1337 South 46th Street, Building 201 Richmond, CA 94804 (415) 972-3150

"We can evade reality, but we cannot evade the consequences of evading reality." - Ayn Rand

From: D'Almeida, Carolyn

Sent: Friday, July 17, 2020 8:16 AM

To: Davis, Eva <<u>Davis.Eva@epa.gov</u>>; 'Jennings, Eleanor' <<u>Eleanor.Jennings@parsons.com</u>>; Steven Willis <<u>steve@uxopro.com</u>>; 'Wayne Miller' <<u>Miller.Wayne@azdeq.gov</u>>; Dan Pope <<u>DPope@css-inc.com</u>>

Cc: Bo Stewart < Bo@Praxis-Enviro.com>

Subject: RE: Some initial thoughts on Final SIP Reports from Microbial Insights

I'm forwarding you all this list we prepared in May 2017 to facilitate your review of the pilot study report when it came in.

I suggest we schedule a call amongst ourselves to discuss the microbial results before the August meeting. What's your availability the week of August 3?

Carolyn d'Almeida Remedial Project Manager Federal Facilities Branch (SFD 8-1) US EPA Region 9 Laboratory 1337 South 46th Street, Building 201 Richmond, CA 94804 (415) 972-3150

"We can evade reality, but we cannot evade the consequences of evading reality." - Ayn Rand

From: Davis, Eva

Sent: Friday, July 17, 2020 6:19 AM

To: 'Jennings, Eleanor' <<u>Eleanor.Jennings@parsons.com</u>>; Steven Willis <<u>steve@uxopro.com</u>>; 'Wayne Miller' <<u>Miller.Wayne@azdeq.gov</u>>; D'Almeida, Carolyn <<u>dAlmeida.Carolyn@epa.gov</u>>; Dan Pope <<u>DPope@css-inc.com</u>>

Cc: Bo Stewart < Bo@Praxis-Enviro.com>

Subject: RE: Some initial thoughts on Final SIP Reports from Microbial Insights

Hi Eleanor -

I really appreciate your input on these results. As I recall, they chose the 4 wells that they thought had the best chance of showing favorable results for this SIP test. Then they primed the wells by injecting sulfate at critical times, but still the results come back showing at best moderate & stressed populations. What does that say about the status of the rest of the site?

I've had even less opportunity to look at this stuff, but I hope that before next month's BCT call, we can have an agencies call to discuss what this data means – especially in light of the ROD that is in place now, and to discuss what we would like the path forward to be. In preparation for that, is there more data that we need to help evaluate the MI data – geochemical data? The rest of the 8260 data (beyond just benzene)? Maybe we need to request that data from Amec now so that we can look at it as we look at the MI data.

Cathy said that the interim contract -12 to 15 months long - would include the new wells we've been fighting for and VI investigation at the Army building, but seems it has been demonstrated that we also need pumping in the CZ to keep control of the plume going to the NE. other plume expansion in other zones may become evident if all pumping is terminated.

Eva

From: Jennings, Eleanor < Eleanor. Jennings@parsons.com >

Sent: Thursday, July 16, 2020 2:44 PM

To: Steven Willis <<u>steve@uxopro.com</u>>; 'Wayne Miller' <<u>Miller.Wayne@azdeq.gov</u>>; D'Almeida, Carolyn <<u>dAlmeida.Carolyn@epa.gov</u>>; Davis, Eva <<u>Davis.Eva@epa.gov</u>>

Subject: RE: Some initial thoughts on Final SIP Reports from Microbial Insights

Sensitive

The short answer:

Our benzene degraders are stressed about something, likely a nutritional deficit. In the long-term, this could possibly pose problems, but I don't have enough information on hand to answer that right now.

The long answer:

Ah, yeah, the term "slowed growth" is really (!) badly named because it implies that, well, growth rates are slowed. And that's not really accurate.

OK, a little background information:

Most of the time, bacteria capable of performing benzene biodegradation fall into a group of bacteria called "Proteobacteria". Like all microbes, the Proteobacteria have cell membranes that are composed of a double-layer of phospholipid fatty acids (PLFAs), dotted with other things like proteins, some structural components, etc. But there are different types of PLFAs, each doing a slightly different job and each sensitive to different things (think of your local community – you have people but one is a baker, one is a banker, one is a shop owner, a few are dog groomers, some are stay-at-home parents....). Certain PLFA's are really sensitive to when the bacterial cell is feeling "stressed". Now, it can be any kind of stress, and when the cell feels stress, these specific PLFAs undergo a chemical change (think of it as switching from facing to the right to facing to the left). This helps to harden the cell wall just a little bit, giving the cell some extra protection in case the source of stress is from the environment. You can actually quantify these specific PLFAs, and how many are facing to the right versus how many are facing to the left. From this, you create a ratio, and this ratio is the number given on the data sheet.

Now, a whole different set of PLFA's also react (in a similar way) to stress, but ONLY when that stress is because something toxic to the cell is in the environment. The mathematical ratio generated from these PLFAs is reported as "Impermeability".

Thus, if you see a high "slowed growth" (better named "stress") ratio, together with a high impermeability ratio, that's a good indication that the cell is stressed and it's because something in the environment is toxic. However, in our situation, we don't see that ... we just see that they are stressed (and they are really stressed, in some MWs) for some reason but the relatively low impermeability ratio value says that environmental toxicity doesn't seem to be the cause. So what is the cause for our all-important, benzene-degrading Proteobacteria being stressed? I don't know, but normally the most common reason (once a toxicity issue is ruled out) is nutritional deficits. If I had to guess, the bacteria now have plenty of carbon source (benzene) and plenty of sulfate (because that was added) but are likely really depleted in other key nutrients (nitrates, phosphates, etc.). This nutritional deficit could possibly cause issues for ongong benzene degradation, but that's better answered after an assessment of some geochemical data that I don't have on hand right now. But, ideally, we want to see this ratio be really low, but it's not in our case.

So how does this related to "slowed growth"? Well, sometimes, some forms of stress CAN slow bacterial growth. And that's a pretty common thing to explain to people so maybe some lab person just named it that. But not all stressors will result in true, slowed growth rates. It's an unfortunate name that still sticks around in the vernacular, unfortunately.

Sorry for the rambling answer, but hopefully it helps. Please ask any more questions – I'm here to help!.

Eleanor M. Jennings, M.S., PhD

Fellow; Project Principal Scientist - Environmental Microbiology and Biogeochemistry Eleanor.Jennings@Parsons.com

"Safety Isn't Expensive. It's Priceless."

From: Steven Willis < steve@uxopro.com> Sent: Thursday, July 16, 2020 2:40 PM

To: Jennings, Eleanor < Eleanor. Jennings@parsons.com >; 'Wayne Miller' < Miller. Wayne@azdeq.gov >; 'd'Almeida,

Carolyn K.' <dAlmeida.Carolyn@epa.gov>; Davis, Eva <Davis.Eva@epa.gov>

Subject: [EXTERNAL] RE: Some initial thoughts on Final SIP Reports from Microbial Insights

Thanks, Eleanor. Can you expand upon/explain the following statement:

"Proteobacterial population demonstrating high degree of physiological stress via the "slowed growth" parameter"

Steve Willis UXO Pro, Inc. steve@uxopro.com 480-316-3373

Sent from Mail [go.microsoft.com] for Windows 10

From: Jennings, Eleanor

Sent: Thursday, July 16, 2020 11:33 AM

To: 'Wayne Miller'; 'd'Almeida, Carolyn K.'; Davis, Eva; Steven Willis

Subject: Some initial thoughts on Final SIP Reports from Microbial Insights

Sensitive

Below are some draft, initial thoughts having looked at the Microbial Insights data, in advance of today's meeting. Let me know if you have any questions or would like to discuss.

-Eleanor

35 days incubation, MI Identifier = 122RC **All four target MWs were seeded with fresh sulfate immediately before Biotrap deployments

- •Total biomass reported for all four MWs is moderate
- •13C label is found at a moderate level in the biomass of one of the MWs, proving benzene biodegradation and incorporation into biomass
- •13C label is found in CO2 captured from the sampler for one of the MWs, proving full mineralization of benzene
- Proteobacterial population demonstrating high degree of physiological stress via the "slowed growth" parameter

52 days incubation, MI Identifier= 081RD **All four target MWs were seeded with fresh sulfate immediately before Biotrap deployments

- •Total biomass reported for all four MWs is moderate
- •13C label is found at moderate levels in the biomass of all four MWs, proving benzene biodegradation and incorporation into biomass
- 13C label is found in CO2 captured from the samplers for all four MWs, proving full mineralization of benzene
- Proteobacterial population demonstrating high degree of physiological stress via the "slowed growth" parameter

Initial, draft comparison between the two deployment times

•The longer deployment time didn't appear to significantly change the level of biomass that had incorporated the C13 isotopic tag in the UWBZ. In the LSZ, the level of biomass that did incorporate the C13 tag increased by 100-fold.

- •The amount of C13 isotopic tag in biomass increased significantly between 35 and 52 days of incubation
- •The amount of C13 isotopic tag in CO2 increased significantly between 35 and 52 days of incubation, reflecting an increased amount of benzene mineralization
- •The percent of the total population composed of Proteobacteria increased significantly between 35 and 52 days of incubation
- Both incubation times report a Proteobacterial population that is physiologically stressed.
- •The fresh seeding of the specific MWs that were to be targeted for BioTrap assessments, immediately before deployment, has created data that is likely a "best case scenario" in comparison to the actual situation of sulfate being added in one location and then being drawn through an area. Targeting specific MWs for sulfate addition immediately before BioTrap deployment makes it difficult to determine what the microbial population would have been like (how active with benzene biodegradation, how healthy, etc.) had it not occurred.
- •The 52-day incubation period likely better demonstrated the impact of fresh sulfate seeding.
- Available data DOES suggest, however, that sulfate addition does stimulate the in-situ benzene biodegradation.
- •Available data does not immediately indicate how long the impact of sulfate-addition lasts at this site on the indigenous microbial community, or how substantial the true impact will be under true injection situations.

Eleanor M. Jennings, M.S., PhD

Fellow; Project Principal Scientist - Environmental Microbiology and Biogeochemistry <u>Eleanor.Jennings@Parsons.com</u> 202.302.9996

"Safety Isn't Expensive. It's Priceless."

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